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HCl- Etched Steel Fiber for Determination of Phthalates in Water Samples by Solid-Phase Microextraction

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Abstract

In the present work, a stainless steel wire was etched by hydrochloric acid during a chemical etching process. The obtained black layer on the surface of the fiber was used as sorbent for extraction of trace amount of phthalates in the aqueous samples by solid phase microextraction. New fiber efficiency was investigated using a home-made solid-phase microextraction (SPME) device and gas chromatography/flame ionization detection (GC-FID). The effect of the key parameters on extraction and desorption process such as extraction time, extraction temperature, salt addition, desorption time and temperature were optimized. New fiber has easy made process, high thermal and mechanical stability and low-cost. Validation of the method was carried out under the optimized condition and the results showed that calibration curves were linear in the range of 5 to 300 µg L−1 for the target analytes. Limits of detection were ranged between 0.02-1.8 $\mu g\ L^{-1}$ (S/N=3). Intra-day, Inter-day and fiber-to-fiber reproducibilites were investigated in different concentrations (at a concentration of 50 $\mu g L^{-1}$, precisions were between, 2.94-6.15, 6.85-8.13 and 8.90-13.76 respectively).

Keywords: *Solid phase microextraction, Gas chromatography, Phthalate esters, Water pollutants, Chemical etching.*

Introduction

Phthalate esters (PEs) are a group of manufactured chemicals with a large variety of industrial and consumer applications [1]. These compounds primarily are added to plastics as a plasticizer [2]. Phthalates are colorless liquids with low vapor pressure; the ones with higher molar mass have low volatility and are slightly

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soluble in water [3]. Since phthalates are not chemically bonded to the polymer chains, they possibly were leached into the surrounding environment from the plastic containers. They also can be released intothe environment during the production and distribution of these compounds; thus they are found in different environmental matrix such as air [4], water [5, 6], soil [7], sewage [8] and sediment [9].

In recent years, some research articles have demonstrated adverse effects of certain PEs on animals and human. According to their opinion, exposure to PEs can be caused to reproductive malformation [10-13], breast cancer [14], thyroid [15], allergy and asthma in children [16], endocrine disruption [17] and, etc. The US Environmental Protection Agency (EPA), European Union (EU) and several other countries, classified some phthalates as priority pollutants [3, 18, 19]. Due to bio-accumulation and harmful effects of phthalates for human and environment, the presence of these chemicals should be evaluated in various matrixes.

Gas chromatography (GC) [20, 21] and high-performance liquid chromatography (HPLC) [22, 23], commonly used to detect phthalates in water samples. Nevertheless, when the concentration levels are low, a pre-treatment step is usually needed. Liquid-liquid extraction (LLE) and solid phase extraction (SPE) techniques have been widely used for this purpose, but these techniques are tedious and need a large amount of organic solvents, which are dangerous.

Among the pre-concentration methods, solid-phase microextraction (SPME) has received increasing attention and is now widely employed in the detection of a variety of compounds due to its simplicity, sensitivity, free solvent and low risk of losinganalyte during the sample preparation steps [24].

SPME-GC technique based on silica fibers with the commercial coatings such as CW-DVB [25], PDMS–DVB [26] and Polyacrylate (PA) [27] have been used earlier. However, silica fibers are fragile and need for great care. To overcome this problem, in recent years, many researchers have focused on the development of metallic fibers such as stainless steel [28], gold [29], copper [30], aluminum [31], titanium [32], silver [33], platinum [34], iron [35], nickel [36] and NiTi alloy [37].

On the other hand, commercial SPME fiber coatings suffer from some drawbacks, such as instability and swelling in organic solvents, the stripping of the coatings, relatively low recommended operating temperature (generally in the range 200–270°C) [38] and high cost [39]. These common problems that SPME users contend with are recognized by researchers and constitute areas of SPME improvement.

In 2009, Xu and coworkers introduced the hydrofluoric acid etched stainless steel as a new fiber SPME and used it for PAHs extraction [40].This new fiber was used for determination of polyborominateddiphenyls in environmental water samples by Chen and coworkers, too [41]

In the present work a stainless steel wire was etched via chemical etching technique; by using various etchantsolutions and the new fiber was applied to SPME of some PEs from aqueous samples.

Experimental

Chemicals

Di-isobutyl (DiBP), di-butyl (DBP), butyl benzyl (BBP) and bis (2-ethylhexyl) (DEHP) phthalate >97% purify,Hydrochloric acid (37%) and sodium chloride (99%) were obtained from Merck (Germany) and used as received. A stock standard solution of 100 mg L^{-1} was prepared in methanol for DiBP, DBP, BBP and DEHP. Stock solutions were stored in the darkness at 4°C. Working solutions were prepared daily by diluting the stock solution with double distilled water. All the used solvents were analytical or HPLC grade. Before use, all containers and glasswares were decontaminated in an ultrasonic bath with acetone and thoroughly washed with ultra-pure water. The stainless steel wires (34 cm×0.3 mm) were purchased from American Orthodontics (WI, USA)

Apparatus

The chromatographic analysis of PEs was carried out using a Varian CP-3380 gas chromatography system (Varian-USA), equipped with a split/split-less injector, flame ionization detector (FID), and CBP-5 (50m×0.32mm I.D. and 1.2μm film thickness) capillary column (Varian-USA). Thecolumn, injector and detector temperatures were set at300, 280 and 300°C, respectively. Helium (>99.999%) was used as a carrier gas with a constant flow rate of 1.5 mL min^{-1}.

The SPME device was home made. It consisted of a 25 gauge, 9.0cm stainless steel spinal needle (Dr. Japan Co., Tokyo, Japan), housed in a 6.0cm hollow metal tubing for protection, and fitted at one end with a metal fitting containing a piece of rubber septum. A piece of steel wire (17 cm×0. 3 mm) passing through the septum acted as the SPME fiber. One end of the fiber was attached to a cap and 2.0 cm of the other end was etched. A RHB2 (IKA-Germany) hot plate-stirrer was used during the extraction procedure to stir and heating the samples. The characteristics of thewire surface were investigated by using a scanning electron microscope (TESCAN VEGA 2- USA).

Fiber preparation

The new fiber was prepared chemically by using hydrochloric acid as an etchant solution. Etching process was carried out by immersing 2.5 cm length of stainless steel fiber into hydrochloric acid for 10 minutes at 35°C. The surface of wire turned black gradually, while hydrogen bubbles were released around the fiber. After 10 minutes the etched fiber was taken out of etching solution, washed with distilled water and dried at 200°C for 1 hour. Then the fiber inserted into the home-made SPME device and conditioned at 300°C in the GC injector under helium gas for 1 hour.

Prior to etching procedure some modifications on the surface of the fiber are necessary. Presence of contaminates, like remain fingerprint or greasy thin layer can cause two different areas on surface of fiber who disruption etching process. For this purpose, at first, the tip of wire polished with fine sandpaper, then it puts into a mixture of acetone and methanol and sonicated for 15 minutes in an ultrasonic bath.

SPME procedure

For evaluation of the new fiber efficiency, it was used for pre-concentration ofmentioned phthalates by DI-SPME in aqueous samples. Before extraction, the fiber was thermally conditioned in GC injection port at 300°C for 10 min. A 50 μgL^{-1} working solution of the mixture of the PEs in distilled water was prepared from the stock solution on a daily basis. SPME procedure was performed by placing an aliquot 7 mL working solution into 10 mL vial, capped with a septum. Magnetic stirring with a 1-cm long Teflon-coated stirring bar was used to agitate the samples at the highest but constant possible rate of stirring. Figure 1 shows the schematic diagram of the SPME set-up used in the present work.

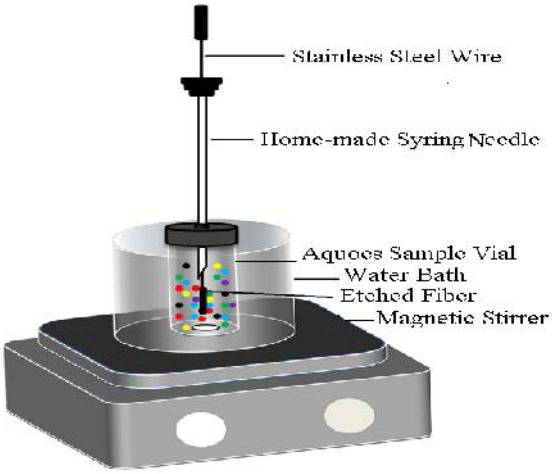

Figure 1. Schematic diagram of the SPME set-up.

The effect of extraction time and temperature, salt addition and desorption time and temperature were investigated.After the extraction, the fiber was withdrawn into the needle and the assembly was pulled out of the extraction vial and after pushing the needle out, it was inserted into the GC injector for separation and chromatographic analysis.

Results and discussion

Fiber optimization

In preliminary experiments, the fiber was prepared by etching with various etchant solutions such as $KMnO_4$/NaOH, $CuSO_4$/ HCl, Picric Acid ($C6H_8O_3$)/H_2SO_4, $Na_2S_2O_5$/ H_2SO_4 and HCl were examined to etching the fibers. The steel wires were immersed to etchant solution for 10 minutes at 35°C and then washed with distilled water and dried at 200°C for 1 hour. Then the fiber inserted into the home-made SPME device and conditioned at 300°C in the GC injector under helium gas for 1 hour. The prepared fibers were used for PEs extraction.As it is obvious in Fig. 2, hydrochloric acid showed the most sensitivity to target phthalates.For this reason, in this study hydrochloric acid selected as theetchant solution.

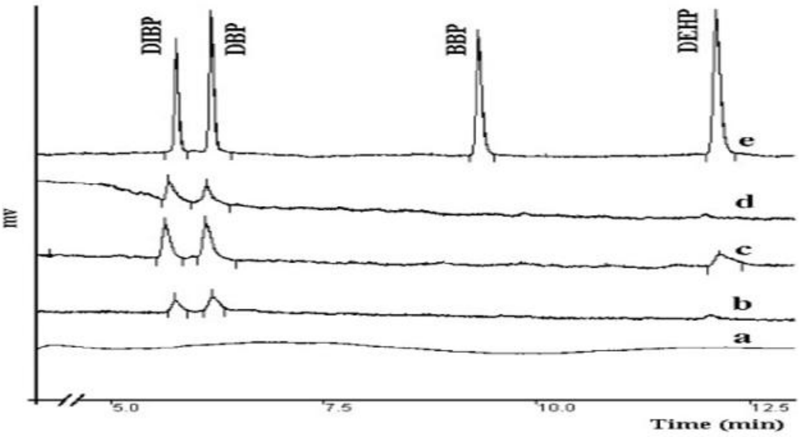

Figure 2. Effect of various etchant solutions on PEs extraction; (a) $KMnO_4$/ NaOH/ H_2O, (b) $CuSO_4$/ HCl/ H_2O, (c) $C_6H_8O_3$/ H_2SO_4, (d) H_2SO_4/ $Na_2S_2O_5$ and (e) HCl.

The surface morphology composition of the steel fiber before and after etching with HCl were analyzed. The SEM images of the steel fiber before and after etching shows that the surface of the surface of the steel fiber was smooth before etching but became rough and porous after etching, that can significantly increase the surface area of the fiber and ensure the sample capacity of the fiber (Figure 3).

SPME optimization

To achieve maximum efficiency of new fiber to PEs extraction from water samples, the effect of various parameters such as extraction time, extraction temperature, ionic strength, desorption time and desorption temperature were studied. The parameters were optimized by one-at-the time process.

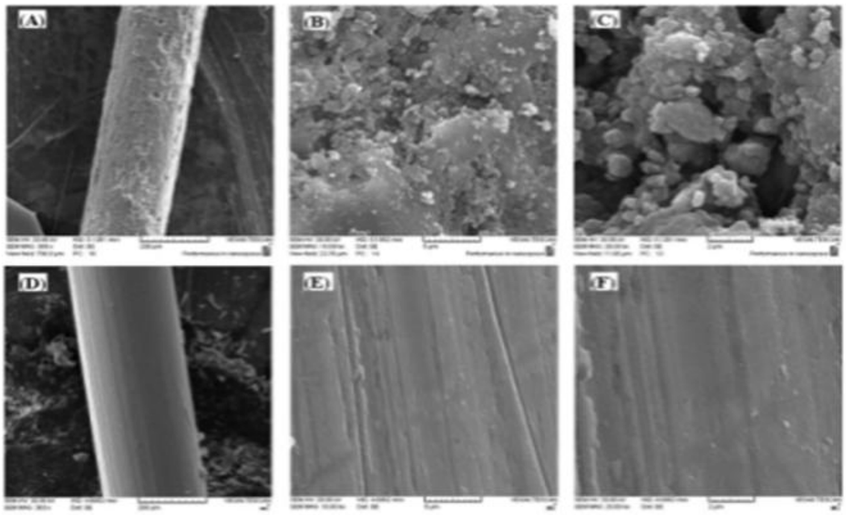

Figure 3. Scanning electron micrographs of the HCl-etched SPME fiber with different magnifications. (A-C): the fiber after etching and (D-F): before etching.

Extraction time

As SPME is an equilibrium-based technique and there is a direct dependence between extraction efficiency and the extraction time, thus it was the first parameter that was optimized. The extraction time was changed from 30 to 60 min. The profiles are shown in Fig.4. As it is clear, for all analytes the extraction efficiency increase up to 50 min and beyond 50 min, the extraction efficiency decreased. Based on the obtained result, 50 min was selected as the optimum extraction time.

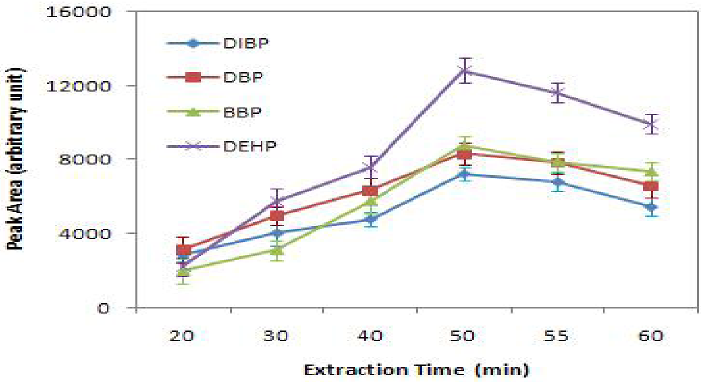

Figure 4. Effect of the extraction time on extraction efficiency of PEs. Conditions: PEs concentration: 50 µgL^{-1}; extraction temperature: 40ºC; desorption temperature: 280ºC and NaCl concentration, 10% (w/v).

Ionic strength and pH

For this purpose usually an inorganic salt is used to enhance ionic strength and mass transfer to the fiber. Therefore effect of this parameter was studied. Experiments were done by adding 0-20% (w/v) NaCl. According to the obtained results (Figure 5), 15% (w/v) showed the best extraction efficiency and was used in the following studied. Due to phthalates are usually stable in a broad range of pH, the effect of this parameter generally was not studied [42].

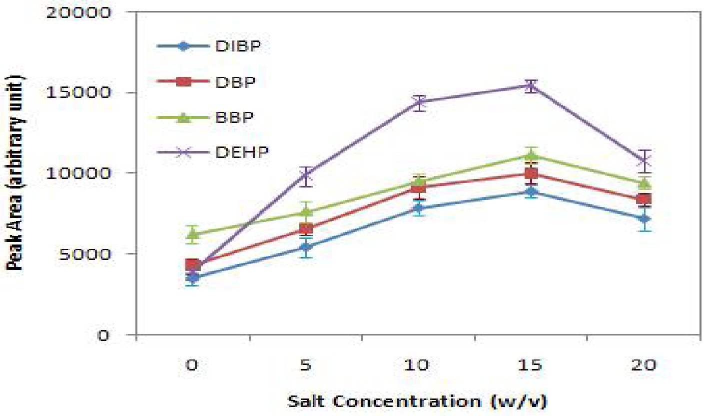

Figure 5. Optimization of ionic strength. Conditions: extraction time: 50 min; PEs concentration: 50 μgL^{-1}; extraction temperature: 40ºC and desorption temperature: 280ºC.

Extraction temperature

The effect of extraction temperature was studied by monitoring chromatographic peak area against extraction temperature. The extraction temperature was changed from 30 to 60°C. An increase in the extraction temperature leads to increasing in diffusion coefficient and a decrease in distribution constant. The amount of extracted PEs increased up to 40°C and then decrease, thus the temperature of 40°C was chosen for subsequent experiments (Figure 6).

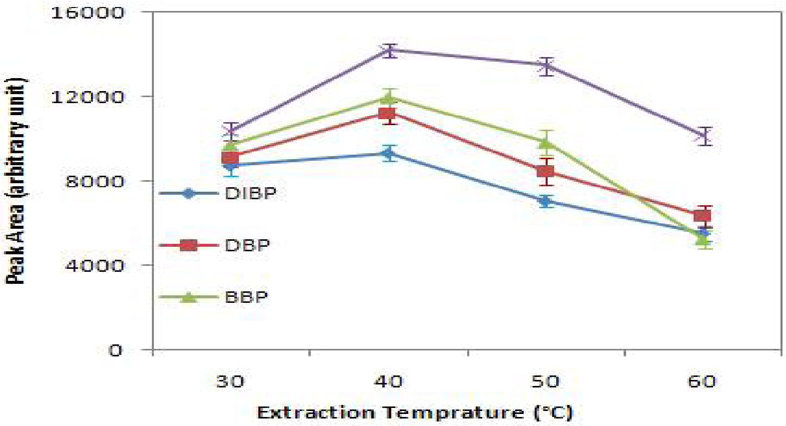

Figure 6. Effect of extraction temperature on the SPME efficiency. Condition: extraction time: 50 min; desorption temperature: 280ºC, and NaCl concentration: 15% (w/v).

Desorption temperature and time

The memory effect is a common problem in determination of phthalates by SPME method. For avoiding carry over and memory effect suitable desorption time and temperature are necessary. For this purpose desorption of the extracted analytes wascarried out at temperatures of 240–300°C.The profiles obtained are shown in Figure 7. As it is obvious the optimal desorption temperature becomesconstant after 280°C.

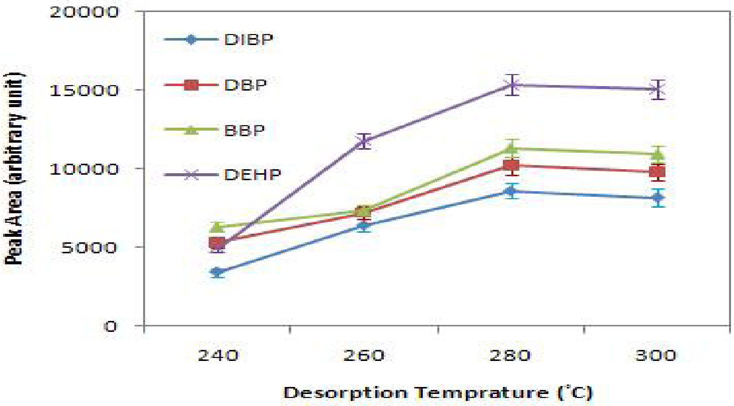

Figure 7. Desorption temperature profiles of PEs. Extraction time, 50 min; Extraction temperature, 40ºC, salt concentration; 15% (w/v); desorption time, 15 min.

Desorptiontimes were also optimized by placing the fiber in the GC injection port for a period 3-15 min. According to the results obtained desorption time set at 12 minutes (Figure 8).

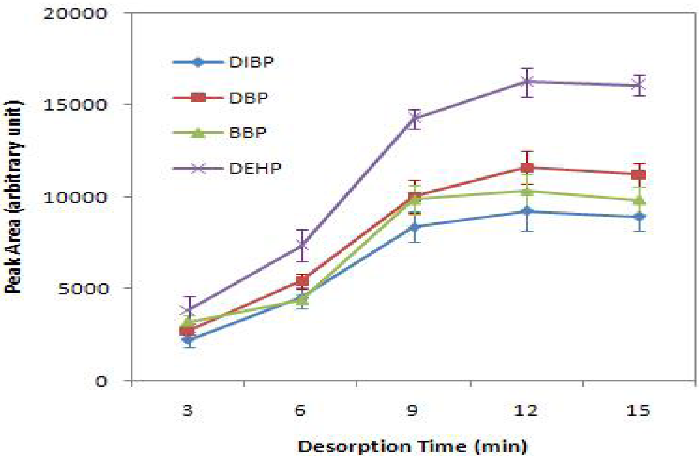

Figure 8. Desorption time profiles of PEs. Extraction time, 50 min; Extraction temperature, 40ºC; salt concentration, 15% (w/v); desorption temperature, 280ºC.

Method validation

Figures of merit including the linear range (LR), repeatability (%R.S.D.) and detection limit (LOD) were evaluated for the proposed work. Table 1 shows some useful analytical data including detection limit (S/N= 3), linear range, recovery, correlation coefficient (r^2) and relative standard deviation (n=3) for desired analytes. The linearity was tested by extracting aqueous solution of the mixture of phthalates using ten concentration range between 1-500 μgL^{-1}. The correlation coefficients (r2) obtained from 0.9975 to 0.9987 and detection limits for studied compounds were between 0.02-1.8

µgL^{-1}. Intra-day, inter-day and fiber-to-fiber reproducibility was investigated in different concentration of 10, 50 and 100 µgL^{-1} of each phthalates. As Table 1, shows intra-day relative standard deviation (%RSD) at a concentration of 10 µgL^{-1} ranged between 2.99-8.00%, while inter-day %RSD was between 3.65-9.95%. Also the fiber-to-fiber reproducibility varied between 8.90-10.68% at a concentration of 50 µgL^{-1}. The validation parameters obtained here are comparable or better than the values reported recently by other groups working with other fibers and using similar techniques (Table 2).

Table 1. Linear range (LR, µg L^{-1}), linearity (r^2), detection limit (LOD, µg L^{-1}), recovery (R) and intra and inter-day precisions (%RSD) of the present DI-SPME-GC method.

Compound	LR ^{a} (µgL^{-1})	r^2	LOD (µgL^{-1})	R±%R.S.D (%)	RSD (%), (n=3)						
					Intra-day			Inter-day			Fiber-to-Fiber
					10	50	100	10	50	100	50 (µg L^{-1})
DiBP	5-100	0.9975	1.8	102±6.2	6.07	6.15	4.59	7.88	6.85	4.96	10.16
DBP	5-100	0.9985	1.5	105±8.3	8.00	6.13	3.89	9.95	7.49	5.17	10.68
BBP	5-100	0.9984	0.020	99±5.5	3.66	5.19	2.96	4.70	8.31	2.81	8.90
DEHP	10-300	0.9987	0.080	97±2.8	2.99	2.94	3.55	3.65	6.88	2.36	13.76

Table 2. Comparison between linear rang, limit of detection and reputability of the present work with similar resul reported by other research groups.

Compounds	References						
	Present work	[25]	[26]	[27]	[42]	[43]	[44]
	HCl-Etched fiber	CW-DVB/ GC-MS	PA/ GC-MS	Calix[4]/ GC-FID	Polyaniline/ GC- FID	CNT-PPY/ GC-FID	Nano-TiO$_2$/ GC-FID
DiBP	5-100 ^{a,b}	-	-	-	-	0.5-300	0.5-1000
	1.8	-	-	-	-	0.1	0.12
	6.15	-	-	-	-	4.1	7.2
DBP	5-100	0.02-10	0.02-10	0.1-100	0.05-150	0.5-300	
	1.5	0.005	0.007	0.006	0.003	0.07	
	6.13	4.4	17	8	7.62	5.1	
BBP	5-100	0.02-10	0.005-10	-	-	0.5-300	0.5-1000
	0.02	0.005	0.02	-	-	0.05	0.09
	5.19	7.4	19	-	-	6.1	7.5
DEHP	10-300	0.05-10	-	0.2-200	-	-	0.5-1000
	0.08	0.04	-	0.010	-	-	0.05
	2.94	28.3	-	9	-	-	6.9

[a] All concentration units: µg L^{-1}

[b] For all compounds; the first, second and third rows of each column are the linear range, detection limit and RSD %, respectively.

Real samples

To apply the proposed method in real sample analysis, two water samples were collected from mineral water bottled (brand A and B), and were then subjected to SPME and GC analysis without any pretreatment. Three replicate analyses were performed on each sample using the HCl-etch fiber at optimal conditions. The result obtained from these analyses are summarized in Table 3 and the obtained chromatograms are shown in Figure 9. In none of the samples DEHP and BBP were detected while DIBP and DBP detected in bothat different concentration levels.

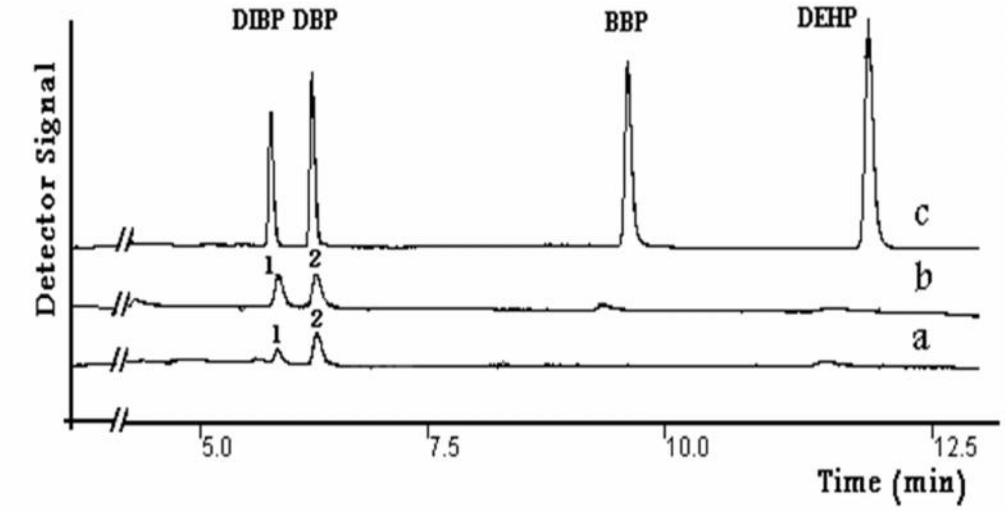

Figure 9. The chromatograms obtained by DI-SPME–GC with HCl-etched fiber for (a) mineral water A, (b) mineral water B and (c) 50 µgL^{-1} standard solution of each PEs.

Table 3. Results obtained for real samples.

Analyte	Mineral water A	Mineral water B
Concentration (µgL^{-1})		
DIBP	8.0	9.6
DBP	11.0	12.0
BBP	N.D	N.D
DEHP	N.D	N.D

Conclusions

In this study a new SPME fiber was prepared by a chemical etching process to monitor phthalates in water samples. The new HCl-etched fiber exhibit porous surface structure and thus high extraction efficiency. The lifetime of the new fiber was such that a single fiber could be used at least100 times for the

DI-SPME analysis of PEs in water, despite the presence of high concentrations of salt in sample solutions during the optimization process and high temperatures used in the desorption step. The method demonstrated suitable precisions, linear ranges, limits of detection and recoveries. New fiber has easy made process, high thermal and mechanical stability and low cost. Overall new fiber has high potential in pre-concentration and determination of phthalate in water samples and it is expected that the steel-HCl-etch fiber has considerable potential for preconcentration and determination of other analysts.

Refrences

[1] J.M.Cano, M.L. Marýn, A. Sanchez, V. Hernandis, *J. Chromatogr. A*, 963, 401(2002).

[2] J. Murphy, Additives for Plastics Handbook. Elsevier Science Ltd, New York(2001).

[3] U.S. Environmental Protection Agency, Hazard Characterization Document, screening-level hazardcharacterization, Phthalate Esters Category, (2010).

[4] A. Turner, M.C. Rawling, *Mar. Chem.*,68, 203 (2000).

[5] A. Peñalver, E. Pocurull, F. Borrull, RM. Marcé, *J. Chromatogr. A*, 922, 377 (2001).

[6] M. Polo, M. Llompart, C. Garcia-Jares, R. Cela,*J. Chromatogr. A*, 1072, 63(2005).

[7] M. Liu, Z. Zeng, H. Fang,*J. Chromatogr. A*, 1076, 16(2005).

[8] F. Jos´eL´opez-Jim´enez, s. Rubio, D.P´erez-Bendito,*Anal. Chim. Acta*, 551, 142(2005).

[9] M.M. Abdeldaiem, J.Rivera-Utrilla, R. Ocampo-Pérez,J.D.Méndez-Díaz,M.J. Sánchez-Polo,*Environ. Manage*, 109, 164(2012).

[10] J. Boberg, S. Christiansen, M.Axelstad, T.S. Kledal, A.M. Vinggaard, M. Dalgaard, C. Nellemann, U. Hass,*Reprod. Toxicol.*, 31, 200(2011).

[11] S.H. Swan, *Environ. Res.*,108, 177(2008).

[12] S.W. Grande, A.J. Andrade, C.E. Talsness, K. Grote, A. Golombiewski, A. Sterner-Kock, I. Chahoud,*Toxicology*, 229, 114(2007).

[13] B.R. Hannas, C.S. Lambright, J. Furr, K.L. Howdeshell, V.S. Wilson, L.E. Gray,*Toxicol. Sci.,* 123, 206(2011).

[14] L. Lopez-Carrillo, R.U. Hernandez-Ramirez, A.M. Calafat, L.Torres-Sanchez, M. Galvan-Portillo, L.L. Needham, R. Ruiz-Ramos, M.E. Cebrián, *Environ. Health Perspect*, 118, 539(2010).

[15] M. Boas, H. Frederiksen, U. Feldt-Rasmussen, N.E. Skakkebaek, L. Hegedus, L. Hilsted, A. Juul, K.M. Main,*Environ Health Perspect*, 118, 1458 (2010).

[16] J.J.K. Jakkola, T.L. Knight,Environ. *Health Perspect*, 116, 845(2008).

[17] M.A. Herreros, A. Gonzalez-Bulnes, C. Letelier, I. Contreras-Solis,J.M. Ros-Rodriguez, T. Encinas, *Theriogenology*, 73, 141 (2010).

[18] EU, Bis(2-ethylhexyl)phthalate (DEHP). EUR 23384 EN/2 (2008)

[19] NRC (National Research Council) The National Academy of Sciences. The Task Ahead (2008)

[20] B. Tienpont, F. David, P. Sandra, F. Vanwalleghem,*J. Microcol*., 4, 194 (2000).

[21] J. Bartulewicz, E. Bartulewicz, J. Gawlowski, J. Niedzielski, *Chem. Anal*., 41, 753 (1996).

[22] M.T. Kelly, M. Larroque,*J. Chromatogr. A*, 841, 177 (1999).

[23] S. Jara, C. Lysebo, T. Greinbrokk, E. Lundanes,*Anal. Chim. Acta*, 407, 165 (2000).

[24] J. Pawliszn, Solid-phase microextraction, theory and practice, Wiley, New York,USA (1996)

[25] K. Luks-Betlej, P. Popp, B. Janoszka, H. Paschke,*J. Chromatogr*. *A*, 938, 93 (2001).

[26] A. Peñalver, E. Pocurull, F. Borrull, R.M. Marcé, *J. Chromatogr. A*, 872, 191(2000).

[27] X. Li, Z. Zeng, Y. Chen, Y. Xu, *Talanta*, 63, 1013(2004).

[28] A. Mollahosseini, E. Noroozian,*Anal. Chim. Acta*, 638, 169(2009).

[29]Dj. Djozan, S. Bahar,*Chromatographia*,59, 95(2004).

[30] Dj. Djozan, Y. Assadi, G. Karim-Nezhad,*Chromatographia*, 56, 611 (2002).

[31] Dj. Djozan, Y. Assadi, S. Hosseinzadeh Haddadi, *Anal. Chem*., 73, 4054(2001).

[32] A. Es-haghi, S.M. Hosseini, Z. Monsef Khoshhesab,*Anal. Chim. Acta*, 742, 74 (2012).

[33] J. Feng J, M. Sun, J. Li, X. Liu, S. Jiang, *Anal. Chim. Acta*, 701 (2011) 174-180.

[34] A. Mohammadi, Y. Yamini, J. Alizadeh,*J. Chromatogr. A*, 1063, 1 (2005).

[35] A. Paszternak, S. Stichleutner, I. Felhosi, Z.Keresztes, F. Nagy, F. Kuzmann, A. Vertes, Z. Homonnay, G. Peto, E. Kalman, *Electrochim. Acta*, 53, 337 (2007).

[36] Dj. Djozan, M.H. Pournaghi-Azar, S. Bahar, *Chromatographia*, 59, 595 (2004).

[37] D. Budziak, E. Martendal, E. Carasek, *J. Chromatogr*. A, 1164, 18 (2007).

[38] Supelco Corp, Manufacturer Data Sheet, Bellefonte, PA, (1996).

[39] A. Kumar Gaurav, A. Kumar Malik, D. Kumar Tewary, B. Singh, *Anal. Chim. Acta*, 610, 1 (2008).

[40] H.L. Xu, Y. Li, D.Q. Jiang, X.P. Yan,*Anal. Chem.,* 81, 4971 (2009).

[41] X.F. Chen, C.G. Cheng, X. Wang, R.S. Zhao,*Anal. Methods*, 4, 2908(2012).

[42] X. Li, M. Zhong, S. Xu, C. Sun,*J. Chromatogr. A*, 1135, 101(2006).

[43] H. Asadollahzadeh, E. Noroozian, Sh. Maghsoudi,*Anal. Chim. Acta*, 669, 32(2010).

[44] M.H. Banitaba, S.S, Hosseiny Davarani, A. Pourahadi, *J. Chromatogr*. A, 1283, 1(2013)..